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*Glomus mosseae*, endomycorrhizal with *Liquidambar styraciflua* L. seedlings retards NO$_3$, NO$_2$ and NH$_4$ nitrogen loss from a temperate forest soil

**Summary**

The influence of a mycorrhizal fungus on downward movement of NH$_4$, NO$_2$, and NO$_3$ nitrogen in forest soil was determined by establishing combinations of soil, fungus and seedlings in plastic pipes and monitoring the nitrogen content of water percolating to two depths. Compared with controls of soil alone and of soil + seedling alone, treatments containing the mycorrhizae showed a significant reduction of NH$_4$-N loss from 5 and 25 cm depths and significant reduction of NO$_3$-N loss from the 5 cm depth. No significant effect was observed on nitrite loss.

**Introduction**

Mycorrhizal development on roots often promote increased nutrient uptake and growth of host trees. However, we are not aware of studies reporting the effect of mycorrhizae on the downward movement of nitrogen in soil profiles. Indeed, Bowen stated, 'Interception of leached nutrients in the soil and litter by hyphae has yet to be measured, but it is unlikely to be by any means complete'.

The present study was designed to determine the effect of *Glomus mosseae* (Nicol. & Gerd.) Gerd. & Trappe, endomycorrhizal with *Liquidambar styraciflua* L. seedlings on the movement of nutrient through soil profiles from the U.S. Forest Service Coweeta Experimental Watershed at Franklin, North Carolina.

**Methods**

Soil from A, B, and C horizons was placed in 15 cm diameter polyvinylchloride pipes. Sterilization to kill indigenous mycorrhizal fungi was performed by heating soil to 90° C for 3 eight-hour periods with steam. All soils were planted with *L. styraciflua* seeds which had been surface sterilized for 30 seconds with 30% H$_2$O$_2$. After establishment, seedlings were pruned back to the soil surface to leave three shoots per pot in some treatments and none in other treatments. Pruned back seedlings were maintained in some treatments to develop some root channels in the soil and in others to support the fungus while minimizing the seedling effect. About 5 g of sandy soil from pots...
planted to *Sorghum* sp. and infested with *G. mossea* were placed in certain treatments as a fungal inoculum. The filtrate from a suspension of the forest soil was added to one treatment to restore some of the indigenous microorganisms which might have been destroyed by the steam treatment. Ceramic water collection cones 1 cm diam. and 10 cm long (Soilmoisture Equip. Corp., Santa Barbara, Calif., U.S.A.), were inserted horizontally through the pipe walls into soil at 5, and 25 cm depths below the soils surface. Each cone was attached to a rigid polyethylene collection bottle which was evacuated to $-0.1$ bar by means of a cartesian diver pressure-vacuum regulator and a water pump aspirator. The experiment was designed to determine the effects of both soil-tree-fungus treatments and nutrient addition treatments on nutrient movement in forest soil columns.

The soil-tree-fungus treatments consisted of A) seedlings pruned back, B) seedlings alone, C) seedlings infested with *G. mossea* but pruned back, D) seedlings infested with *G. mossea*, and E) seedlings infested with *G. mossea* and inoculated with filtrate of forest soil extract. Each of the five treatments was replicated eight times all on one greenhouse bench.

Nutrient treatments were designed 1) to supply Na, K, Mg, and Ca at concentrations found in rain water reaching the forest floor at Coweeta where the soils originated, 2) to provide equal amounts of nitrogen first as NH$_4$-N, then as NO$_3$-N, and finally 3) to overcome an apparent phosphorus deficiency in the *L. styraciflua*. The NH$_4$-N treatment solution consisted of 41.25 mg (NH$_4$)$_2$HPO$_4$/l. The NO$_3$-N containing solutions was comprised of 206.4 mg CaSO$_4$.7H$_2$O, 107.4 mg MgCl.6H$_2$O, 53 mg NaNO$_3$, and 114.6 mg KCl/l. Both solutions thus contained 8.7 mg nitrogen/l. The third solution was deionized water. On a given date all pipes received 500 ml aliquots of the same nutrient solution. After 24 hours, percolates were collected for analyses. Several days later when soil surfaces appeared dry, 500 ml aliquotes of a different solution were applied to each pipe and new percolates collected. The order of solution application was 1) deionized water, 2) the NH$_4$-N containing solution, 3) deionized water, 4) the NO$_3$-N containing solution, and 5) deionized water.

Concentrations of NH$_4$, NO$_3$, and NO$_2$ nitrogen as well as PO$_4$-P in collected percolate were determined colorimetrically. Ion concentrations were not corrected for differences either in tree seedling dry weight or relative mycorrhizal infestation because plants are still in use.

**Results**

Analysis of variance showed highly significant ($p < .001$) soil-tree-fungus treatment effects and nutrient treatment effects for all ions at all depths except for NO$_2$-N. The soil-tree-fungus effect on NO$_2$-N was significant at $p < .025$. Student-Newman-Keuls *a posteriori* multiple range tests were applied to determine which means differed significantly. Mean nutrient concentrations at two sampling depths and results of multiple range tests are shown in Table 1. Each mean represents the nutrient concentration from the 8 replicate plastic pipes in each of the soil-tree-fungus treatments averaged over all the 5 nutrient treatments. There were some significant interactions effects between the two types of treatments. Details of nutrient treatment
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TABLE 1

Ion concentrations in mg/liter, for five soil-tree-fungus treatments. (See text for treatment details). Means not significantly different at \( P < .01 \) are connected by solid line while those not different at \( P < .05 \) are connected by a broken line, \( N = 40 \) except for \( \text{NO}_2 \) at 25 cm where for C, D, and E it was 39, 35, and 35.

<table>
<thead>
<tr>
<th>Depth</th>
<th>A. soil</th>
<th>B. soil</th>
<th>C. soil</th>
<th>D. soil</th>
<th>E. soil</th>
</tr>
</thead>
<tbody>
<tr>
<td>05 cm</td>
<td>1.38</td>
<td>1.35</td>
<td>.307</td>
<td>.558</td>
<td>1.15</td>
</tr>
<tr>
<td>25 cm</td>
<td>4.70</td>
<td>4.14</td>
<td>.655</td>
<td>.674</td>
<td>.363</td>
</tr>
<tr>
<td>05 cm</td>
<td>25.6</td>
<td>31.1</td>
<td>2.73</td>
<td>.979</td>
<td>.076</td>
</tr>
<tr>
<td>25 cm</td>
<td>124.</td>
<td>119.</td>
<td>92.5</td>
<td>45.4</td>
<td>12.8</td>
</tr>
<tr>
<td>05 cm</td>
<td>.159</td>
<td>.136</td>
<td>.034</td>
<td>.007</td>
<td>.004</td>
</tr>
<tr>
<td>25 cm</td>
<td>.09</td>
<td>.138</td>
<td>.111</td>
<td>.037</td>
<td>.048</td>
</tr>
</tbody>
</table>

Effects and interactions will be reported elsewhere after plants have been harvested and relative mycorrhizal infestation is determined.

Discussion

The endomycorrhizal symbiosis clearly reduced the loss of \( \text{NH}_4 \) and \( \text{NO}_3-N \) compared to losses observed from soil columns alone and from soil columns containing \( L. \text{styraciflua} \) alone. The sources of \( \text{NH}_4 \) and \( \text{NO}_3-N \) nitrogen are less clear. The concentrations of \( \text{NH}_4 \) and \( \text{NO}_3-N \) in the nutrient application treatments were 8.7 mg nitrogen per liter. Ammonia concentrations in the percolate were consistently less than those applied, but they increased with depth.

Nitrate-N in percolates from the soil alone and from soil supporting \( L. \text{styraciflua} \) alone obtained at the 5 cm depth were three times greater, while those from 25 cm depth were about fourteen times greater than concentrations in the applied solution. This suggests that most of the \( \text{NO}_3 \) moving through the soil columns came from mineralization of soil organic matter. These high background \( \text{NO}_3-N \) concentrations at the 25 cm depth can probably account for the reduced significance of differences between \( \text{NO}_3 \) concentrations in percolates of the mycorrhizal and non-mycorrhizal treatments. If the ambient \( \text{NO}_3-N \) concentrations exceed the concentration at which maximum \( \text{NO}_3-N \) uptake rate occurs, the reduction of soil solution \( \text{NO}_3-N \) concentrations could not be as great as at lower ambient concentrations. The concentrations at which maximum or half of the maximum \( \text{NO}_3-N \) uptake velocity occur in this endomycorrhizal system are not known. The magnitudes...
of these concentrations can probably be estimated from results of the one
NO₃-N uptake study judged reliable by Epstein in which half of the
maximum uptake rate was reached in Zea mays at 0.28 mg NO₃-N/l and the
maximum concentration used in the study was 3.3 mg NO₃-N/l. In the
present study the 25 cm depth background NOg-N concentration was 37
times higher than the maximum used with Zea mays and 425 times greater
than the concentration at which half of the maximum uptake was observed.

The mineralization of nitrogen containing organic matter to NH₄ and its
subsequent oxidation to NO₃ may account for the relatively high losses of
NO₃. This apparent NH₄ to NO₃ conversion or nitrification is surprising be-
cause there is mounting evidence that climax ecosystems inhibit nitrifica-
tion. Both the soil and L. styraciflua were from climax oak-hickory forests.
Any inhibitors of nitrification if originally present must have been degraded
by the time studies of nitrogen movement had begun.

Even greater reduction of nitrogen loss by the tree–fungus symbiosis treat-
ments might have occurred if plant growth had been limited by the soil
nitrogen supply. Instead, L. styraciflua growth appeared to be phosphorus
limited. The plants in all treatments showed classic signs of phosphorus de-
ficiency — purple veins and blotches on leaves. Also dissolved phosphorus was
detectable only at the 5 cm depth only on the day following phosphorus
application.

The results of this study show that mycorrhizal roots significantly reduce
the soil solution concentrations of both NH₄ and NO₃-N compared to non-
mycorrhizal controls. Although actual net water flow rates were not measured
in the soil columns, flow rates in columns containing transpiring plants might
equal but could not exceed flow rates in columns having the small non-
mycorrhizal plants or lacking plants. Under a regime of equality of flow rates
in all treatments or reduced flow rates in pipes containing the mycorrhizal
symbionts, the significant reduction of soil solution nitrate and ammonia
concentrations by the symbionts means that the symbionts significantly re-
duced the net nitrogen loss from the soil columns. The endomycorrhizal sym-
biosis is therefore ecologically significant as a nitrogen conserver.

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